7. A method of cleaving a polynucleotide, comprising the steps of:

(a) contacting a sample suspected of containing a target nucleic acid of interest, said target nucleic acid comprising a first portion and a second portion located immediately 3' to the first portion, with:

(i) a 5'-polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region that is capable of specifically hybridizing to the first portion of the target nucleic acid and a 5'-region located immediately 5' to the 3'-region; and

(ii) a 3'-polynucleotide probe comprising a 5'-region that is capable of specifically hybridizing to the second portion of the target nucleic acid and a 3'-region located immediately 3' to the 5'-region,

under conditions in which the 3'-region of the 5'-probe and the 5'-region of the 3'-probe specifically hybridize immediately contiguously with one another to the first and second portions, respectively, of the same target nucleic acid molecule;

(b) selectively cleaving the 5'-polynucleotide probe to release a nucleotide or a polynucleotide from its 5'-region; and

(c) detecting and/or quantifying said cleavage.

21. A method of detecting the presence of a target nucleic acid in a sample, comprising the steps of

(a) contacting a sample suspected of containing a target nucleic acid of interest with:

(i) a FEN-1 polypeptide;

(ii) 5'-polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region that is capable of specifically hybridizing to a first portion of the target nucleic acid and a 5'-region located immediately 5' to the 3'-region; and

(iii) a 3'-polynucleotide probe comprising a 5'-region that is capable of specifically hybridizing to a second portion of the target nucleic acid which is located immediately 3' to the first portion and a 3'-region located immediately 3' to the 5'-region,

under conditions in which the 3'-region of the 5'-probe and the 5'-region of the 3'-probe specifically hybridize immediately contiguously with one another to the first and second portions, respectively, of the same target nucleic acid molecule; and

(b) detecting the presence or absence of, and/or quantifying the amount of, FEN-1 polypeptide-generated cleavage, thereby detecting the presence of the target nucleic acid in the sample.

36. A method of detecting the presence of a target nucleic acid in a sample, comprising the steps of:

(a) contacting a sample suspected of containing a target nucleic acid of interest with:

(i) a FEN-1 polypeptide;

(ii) 5'-polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region that is capable of specifically hybridizing to a first portion of the target nucleic acid and a 5'-region located immediately 5' to the 3'-region; and

(iii) a 3'-polynucleotide probe comprising a 5'-region that is capable of specifically hybridizing to a second portion of the target nucleic acid which is located immediately 3' to the first portion and a 3'-region located immediately 3' to the 5'-region,

under conditions in which the 3'-region of the 5'-probe and the 5'-region of the 3'-probe specifically hybridize immediately contiguously with one another to the first and second portions, respectively, of the same target nucleic acid molecule to form a structure that the FEN-1 polypeptide is capable of binding, thereby yielding a target-FEN-1 complex; and

(b) detecting the presence or absence of the target-FEN-1 complex, thereby detecting the presence of the target nucleic acid in the sample.

51. A hybridization complex comprising:

(a) a bridge polynucleotide comprising a first portion and second portion located immediately 3' to the first portion;

(b) a first polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region and a 5'-region located immediately 5' to the 3'-region; and

(c) a second polynucleotide probe comprising a 5'-region and a 3'-region located immediately 3' to the 5'-region,

wherein the 3'-region of the first probe and the 5'-region of the second probe are specifically hybridized immediately contiguously with one another to the first and second portions, respectively, of the same bridge polynucleotide molecule, thereby forming a hybridization complex.

EXHIBIT B

Copy of Pending Claims Upon Entry of this Amendment

1. An isolated polynucleotide encoding a FEN-1 polypeptide as shown in SEQ ID NO:1 or SEQ ID NO:3, or a fragment of said polypeptide having flap endonucleolytic cleavage activity.

2. An isolated polynucleotide, wherein said polynucleotide comprises a nucleotide sequence selected from the group consisting of SEQ ID NOS:29-51.

3. An isolated polynucleotide of Claim 2, wherein said polynucleotide comprises the sequence of SEQ ID NO:28.

4. A host cell comprising the polynucleotide of Claim 1.

5. A non-mammalian host cell comprising a mammalian FEN-1 polypeptide of Claim 1.

6. The poly nucleotide of Claim 1 that is full length.

7. A method of cleaving a polynucleotide, comprising the steps of:

(a) contacting a sample suspected of containing a target nucleic acid of interest, said target nucleic acid comprising a first portion and a second portion located immediately 3' to the first portion, with:

(i) a 5-polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region that is capable of specifically hybridizing to the first portion of the target nucleic acid and a 5'-region located immediately 5' to the 3'-region; and

(ii) a 3'-polynucleotide probe comprising a 5'-region that is capable of specifically hybridizing to the second portion of the target nucleic acid and a 3'-region located immediately 3' to the 5'-region,

under conditions in which the 3'-region of the 5'-probe and the 5'-region of the 3'-probe specifically hybridize immediately contiguously with one another to the first and second portions, respectively, of the same target nucleic acid molecule;

(b) selectively cleaving the 5'-polynucleotide probe to release a nucleotide or a polynucleotide from its 5'-region; and

(c) detecting and/or quantifying said cleavage.

8. The method of Claim 7 in which step (c) comprises detecting the presence of the released nucleotide or polynucleotide, where the presence of the released nucleotide or polynucleotide correlates with the presence of the target nucleic acid in the sample.

9. The method of Claim 7 in which step (c) comprises quantifying the amount of nucleotide or polynucleotide released, where the quantity of released nucleotide or polynucleotide correlates with the presence or abundance of the target nucleic acid in the sample.

10. The method of Claim 7 in which the selective cleavage is accomplished using a yeast FEN-1 polypeptide or a mammalian FEN-1 polypeptide.

11. The method of Claim 10 in which the FEN-1 polypeptide is a human FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:1 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

12. The method of Claim 10 in which the FEN-1 polypeptide is a murine FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:3 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

13. The method of Claim 10 in which the FEN-1 polypeptide is a yeast FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:5 or SEQ ID NO:7 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

14. The method of Claim 7 in which the 3'-probe comprises a 3'-flap region that is 1 to 10 nucleotides in length.

15. The method of Claim 14 in which the 3'-flap region is 1 nucleotide in length.

16. The method of Claim 7 in which the 5'-probe comprises a 5'-flap region that is 1 to 20 nucleotides in length.

17. The method of Claim 7 in which the 5'-probe contains a detectable label.

18. The method of Claim 17 in which the 5'-probe comprises a 5'-flap region that contains a detectable label.

19. The method of Claim 18 in which the 5'-end of the 5'-probe contains a detectable label.

20. The method of Claim 7 in which the 5'-probe is immobilized on a substrate.

21. A method of detecting the presence of a target nucleic acid in a sample, comprising the steps of:

(a) contacting a sample suspected of containing a target nucleic acid of interest with:

(i) a FEN-1 polypeptide;

(ii) a 5-polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region that is capable of specifically hybridizing to a first portion of the target nucleic acid and a 5'-region located immediately 5' to the 3'-region; and

(iii) a 3'-polynucleotide probe comprising a 5'-region that is capable of specifically hybridizing to a second portion of the target nucleic acid which is located immediately 3' to the first portion and a 3'-region located immediately 3' to the 5'-region,

under conditions in which the 3'-region of the 5'-probe and the 5'-region of the 3'-probe specifically hybridize immediately contiguously with one another to the first and second portions, respectively, of the same target nucleic acid molecule; and

(b) detecting the presence or absence of, and/or quantifying the amount of, FEN-1 polypeptide-generated cleavage, thereby detecting the presence of the target nucleic acid in the sample.

22. The method of Claim 21 in which the 5'-probe contains a detectable label.

23. The method of Claim 22 in which the 5'-probe comprises a 5'-flap region that contains a detectable label.

24. The method of Claim 23 in which the 5'-end of the 5'-probe contains a detectable label.

25. The method of Claim 21 in which the 5'-probe is immobilized on a support.

26. The method of Claim 21 in which the FEN-1 polypeptide is a mammalian or a yeast FEN-1 polypeptide.

27. The method of Claim 26 in which the FEN-1 polypeptide is a yeast FEN-1 polypeptide, a murine FEN-1 polypeptide or a human FEN-1 polypeptide.

28. The method of Claim 26 in which the FEN-1 polypeptide is a human FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:1 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

29. The method of Claim 26 in which the FEN-1 polypeptide is a murine FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:3 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

30. The method of Claim 26 in which the FEN-1 polypeptide is a yeast FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:5 or SEQ ID NO:7 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

31. The method of Claim 21 in which the 3'-probe comprises a 3'-flap region that is 1 to 10 nucleotides in length.

32. The method of Claim 21 in which the 3'-flap region is 1 nucleotide in length.

33. The method of Claim 21 in which the 5'-probe comprises a 5'-flap region that is 1 to 20 nucleotides in length.

34. The method of any one of Claims 21-33 in which the amount of FEN-1 polypeptide-generated cleavage is quantified.

35. The method of any one of Claims 21-33 in which the presence or absence of FEN-1 polypeptide-generated cleavage is detected.

36. A method of detecting the presence of a target nucleic acid in a sample, comprising the steps of:

(a) contacting a sample suspected of containing a target nucleic acid of interest with:

(i) a FEN-1 polypeptide;

(ii) a 5-polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region that is capable of specifically hybridizing to a first portion of the target nucleic acid and a 5'-region located immediately 5' to the 3'-region; and

(iii) a 3'-polynucleotide probe comprising a 5'-region that is capable of specifically hybridizing to a second portion of the target nucleic acid which is located immediately 3' to the first portion and a 3'-region located immediately 3' to the 5'-region,

under conditions in which the 3'-region of the 5'-probe and the 5'-region of the 3'-probe specifically hybridize immediately contiguously with one another to the first and second portions, respectively, of the same target nucleic acid molecule to form a structure that the FEN-1 polypeptide is capable of binding, thereby yielding a target-FEN-1 complex; and

(b) detecting the presence or absence of the target-FEN-1 complex, thereby detecting the presence of the target nucleic acid in the sample.

37. The method of Claim 36 in which the FEN-1 polypeptide contains a detectable label.

38. The method of Claim 36 in which the 5'-probe is immobilized on a substrate.

39. The method of Claim 36 which is performed in the absence of Mg^{2+}.

40. The method of Claim 36 in which the 5'-probe contains a detectable label.

41. The method of Claim 40 in which the 5'-probe comprises a 5'-flap region that contains a detectable label.

42. The method of Claim 41 in which the 5'-end of the 5'-probe contains a detectable label.

43. The method of Claim 36 in which the FEN-1 polypeptide is a mammalian or a yeast FEN-1 polypeptide.

44. The method of Claim 43 in which the FEN-1 polypeptide is a yeast FEN-1 polypeptide, a murine FEN-1 polypeptide or a human FEN-1 polypeptide.

45. The method of Claim 43 in which the FEN-1 polypeptide is a human FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:1 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

46. The method of Claim 43 in which the FEN-1 polypeptide is a murine FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:3 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

47. The method of Claim 43 in which the FEN-1 polypeptide is a yeast FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:5 or SEQ ID NO:7 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

48. The method of Claim 36 in which the 3'-probe comprises a 3'-flap region that is 1 to 10 nucleotides in length.

49. The method of Claim 36 in which the 3'-flap region is 1 nucleotide in length.

50. The method of Claim 36 in which the 5'-probe comprises a 5'-flap region that is 1 to 20 nucleotides in length.

51. A hybridization complex comprising:

(a) a bridge polynucleotide comprising a first portion and second portion located immediately 3' to the first portion;

(b) a first polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region and a 5'-region located immediately 5' to the 3'-region; and

(c) a second polynucleotide probe comprising a 5'-region and a 3'-region located immediately 3' to the 5'-region,

wherein the 3'-region of the first probe and the 5'-region of the second probe are specifically hybridized immediately contiguously with one another to the first and second portions, respectively, of the same bridge polynucleotide molecule, thereby forming a hybridization complex.

52. The hybridization complex of Claim 51 in which the first probe contains a detectable label.

53. The hybridization complex of Claim 52 in which the 5'-region of the first probe contains a detectable label..

54. The hybridization complex of Claim 53 in which the 5'-end of the first probe contains a detectable label.

55. The hybridization complex of Claim 51 in which the first probe is immobilized on a substrate.

56. The hybridization complex of Claim 51 in which the second probe comprises a 3'-flap region that is 1 to 10 nucleotides in length.

57. The hybridization complex of Claim 56 in which the 3'-flap region is 1 nucleotide in length.

58. The hybridization complex of Claim 51 in which the first probe comprises a 5'-flap region that is 1 to 20 nucleotides in length.

59. A kit for detecting the presence of a target nucleic acid in a sample, comprising:

(a) a FEN-1 polypeptide;

(b) a first polynucleotide probe capable of being cleaved by a FEN-1 polypeptide, comprising a 3'-region capable of specifically hybridizing to a first portion of a target nucleic acid of interest and a 5'-region located immediately 5' to the 3'-region; and

(c) a second polynucleotide probe comprising a 5'-region capable of specifically hybridizing to a second portion of the target nucleic acid which is located immediately 3' to the first portion and a 3'-region located immediately 3' to the 5'-region,

wherein the 3'-region of the first probe and the 5'-region of the second probe are capable of specifically hybridizing immediately contiguously with one another to the first and second portions, respectively, of the same target nucleic acid molecule to form a structure that is capable of being bound or cleaved by the FEN-1 polypeptide.

60. The kit of Claim 59 further in which the first or second probe contains a detectable label.

61. The kit of Claim 59 in which the FEN-1 polypeptide contains a detectable label.

62. The kit of Claim 59 in which the second probe comprises a 3'-flap region that is 1 to 10 nucleotides in length.

63. The kit of Claim 59 in which the 3'-flap region is 1 nucleotide in length.

64. The kit of Claim 59 in which the first probe comprises a 5'-flap region that is 1 to 20 nucleotides in length.

65. The kit of Claim 59 in which the first probe contains a detectable label.

66. The kit of Claim 59 in which the first probe comprises a 5'-flap region that contains a detectable label.

67. The kit of Claim 66 in which the 5'-end of the first probe contains a detectable label.

68. The kit of Claim 59 in which the first or second probe is immobilized on a substrate.

69. The kit of any one of Claim 59-68 in which the FEN-1 polypeptide is a mammalian or a yeast FEN-1 polypeptide.

70. The kit of Claim 69 in which the FEN-1 polypeptide is a yeast FEN-1 polypeptide, a murine FEN-1 polypeptide or a human FEN-1 polypeptide.

71. The kit of Claim 70 in which the FEN-1 polypeptide is a human FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:1or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

72. The kit of Claim 70 in which the FEN-1 polypeptide is a murine FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:3 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.

73. The kit of Claim 70 in which the FEN-1 polypeptide is a yeast FEN-1 polypeptide comprising the amino acid sequence shown in SEQ ID NO:5 or SEQ ID NO:7 or a fragment thereof having 5'-flap endonucleolytic cleavage activity.